Journal of Biomedical & Laboratory Sciences
Volume 5, Number 2, April, 1993

Analysis of Pterins in Dried Urine Spots Collected on Filter Paper for Detecting Patients with Tetrahydrobiopterin Synthesis Deficiency

TT Liu[1]*, MH Chiang[3], YC Chen[3] and KJ Hsiao[2,3]

Insts. of Biochem[1], and Genetics[2], National Yang-Ming Medical College; Dept. of Med. Res[3], Veterans General Hospital-Taipei

Phenylketonuria (PKU) is an inherited metabolic disorder of phenylalanine hydroxylation. Defects in tetrahydrobiopterin (BH_4), a cofactor required in phenylalanine hydroxylation, or phenylalanine hydroxylase itself may cause variant forms of PKU. Differential diagnosis of variant forms of PKU by urinary pterins analysis had been established in our laboratory in aid of choosing proper treatment. In attempt to spread the differential diagnosis service over remote area, analyzing urine collected on filter paper was studied. Urine collected in ascorbic acid was spotted onto the filter paper and dried in dim light. The dried filter paper can then be send by mail. Neopterin (N) and biopterin (B) in dried urine spot were extracted by water. Reduced form of N and B in the extract were oxidized by MnO_2 in acidic condition. N and B were analyzed by reverse phase (C-18) high performance liquid chromatography with an isocratic elution of 3% methanol and with fluorescent detection (ex. 350 nm, em. 450 nm). Within-run and run-to-run imprecision (C.V. %) of N and B were around 4% and 8%, respectively. The N and B level measured from dried durine spots agreed with that assayed from the same urine specimens of both normal and PKU patients. Using dried urine spot method, urinary N and B of 52 PKU specimens mailed from overseas were analyzed. Among which one PKU was detected as BH_4 synthesis deficiency. These results indicated that the measurement of N and B in dried urine spot might be applied for differential diagnosis of variants of PKU.